CLAIMS

That which is claimed is:

1. Substantially pure DNA encoding a protease-activated receptor 3.

2. The DNA of claim 1, wherein the DNA is mammalian.

3. Substantially pure DNA having the nucleotide sequence selected from the group consisting of Fig. 1 (SEQ ID NO:1), or degenerate variants thereof, and encoding the amino acid sequence of Fig. 1 (SEQ ID NO:3); Fig. 2 (SEQ ID NO:2), or degenerate variants thereof encoding an amino acid sequence comprising the amino acid sequence of Fig. 1 (SEQ ID NO:3); Fig. 3 (SEQ ID NO:4), or degenerate variants thereof encoding the amino acid sequence of Fig. 2 (SEQ ID NO:6); and Fig. 4 (SEQ ID NO:5), or degenerate variants thereof encoding an amino acid sequence comprising the amino acid sequence of Fig. 3 (SEQ ID NO:6).

4. Substantially pure DNA having 50% or greater sequence identity to the DNA sequence of a sequence selected from the group consisting of SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:4 and SEQ ID NO:5 and which hybridizes to the DNA sequence SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, or SEQ ID NO:5, respectively.

5. An isolated protease-activated receptor 3 protein.

6. The substantially pure protein of claim 5 having an amino acid sequence selected from the group consisting of the sequence shown in Fig. 1 (SEQ ID NO:3). and the sequence shown in Fig. 2 (SEQ ID NO:6).

7. A substantially pure polypeptide having an amino acid sequence which is at least 80% identical to an amino acid sequence selected from the group consisting of the sequence shown in Fig. 1 (SEQ ID NO:3) and the sequence shown in Fig. 2 (SEQ ID NO:6), wherein

a) said polypeptide is activated by thrombin; and

b) said polypeptide mediates phosphoinositide hydrolysis in a cell expressing said polypeptide on its surface.

8. A substantially pure polypeptide which is a fragment or analog of a protease-activated receptor 3 comprising a domain capable of activation by thrombin and mediating phosphoinositide hydrolysis.

9. A vector comprising the DNA of claim 1.

10. A cell comprising the vector of claim 9.

11. An assay device, comprising:
a support surface;
and a cell of claim 10.

12. The assay device of claim 11, wherein the cell is bound to the support surface or present in a suspension on the support surface.

13. A method of testing a candidate compound for its ability to act as an agonist of a protease-activated receptor 3 ligand, the method comprising:

a) contacting a candidate compound with a cell which expresses on its surface a recombinant protease-activated receptor 3 protein or biologically active fragment or analog thereof;

b) measuring PAR3-mediated response of the cell; and

c) identifying the candidate compound as an agonist wherein the contacting causes a substantial increase in PAR3-mediated response.

14. A method of testing a candidate compound for the ability to act as an antagonist of a protease-activated receptor 3 ligand, the method comprising:

a) contacting in the presence of a protease-activated receptor agonist a candidate compound with a cell which expresses on its surface a recombinant protease-activated receptor 3 protein or biologically active fragment or analog thereof;

b) measuring PAR3-mediated response of the cell; and

c) identifying the candidate compound as an antagonist wherein the contacting causes a substantial decrease in PAR3-mediated response relative to PAR3-mediated response in the absence of the candidate antagonist.

15. The method of claim 14, wherein the cell is a mammalian cell which normally presents substantially no protease-activated receptor 3 on its surface, the PAR3-mediated response measured in intracellular phosphoinositide hydrolysis in the cell.

16. The method of claim 14, further comprising:

mixing thrombin with platelets and the identified candidate compound; and

observing the effect of the candidate compound on mediating platelet aggregation.

17. A therapeutic composition, comprising:

a protease-activated receptor 3 ligand agonist; and

a physiologically-acceptable carrier.

18. A therapeutic composition, comprising:

a protease-activated receptor 3 ligand antagonist; and

a physiologically-acceptable carrier.

19. The composition of claim 18, wherein the antagonist is selected from the group consisting of:

(1) the isolated sequence LPIKTFRGAPPNSFEEFPFSALE;

(2) uncleavable thrombin inhibitor LPIKPFRGAPPNSFEEFPFSALE where the PAR 3 cleavage site P1' is muted to block cleavage;

(3) uncleavable thrombin inhibitor LP(hR)TFRGAPPNSFEEFPFSALE where the PAR 3 cleavage site P1 is mutated to block cleavage;
hR is beta-homoarginine (the extra methylene group is in the main chain);

(4) uncleavable thrombin inhibitor (dF)PRPFRGAPPNSFEEFPFSALE where the good active site binding sequence dFPR is substituted for LPIK; dF is D-Phenylalanine;

(5) any of (1)-(4) above where all or part of the sequence TFRGAPNS is replaced with spacer sequences such as GGG;

(6) variations and combinations of (1)-(5) which act as antagonists.

20. A method of treatment, comprising:
administering to a patient a therapeutically effective amount of the composition of claim 18.